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EVALUATION OF FUNGICIDES FOR CONTROL OF PHOMOPSIS CANKER OF DOUGLAS-FIR AT HUMBOLDT NURSERY

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ABSTRACT

Sixteen fungicides were evaluated in vitro and five were evaluated in field trials for control of Phomopsis canker of 2-0 Douglas-fir at Humboldt Nursery. Most of the fungicides inhibited or reduced spore germination compared to no treatment. Six fungicides prevented (benomyl, imazalil, prochloraz, diniconazole) or reduced (chlorothalonil, mancozeb) mycelial growth at 1 ppm. Five fungicides (diniconazole, chlorothalonil, benomyl, mancozb, and imazalil) tested in the field gave significantly better control than no treatment. Benomyl, the fungicide registered for Phomopsis control, was more efficacious than mancozeb and imazalil, but was not as effective as diniconazole or chlorothalonil.

INTRODUCTION

Phomopsis canker of 2-0 Douglas-fir, caused by Phomopsis occulta, usually results in scattered and infrequent losses at Humboldt Nursery. About 5% of the crop is lost each year. However, incidence of the disease has increased the last few growing seasons. The pathogen has also caused needle loss and shoot blight of 1-0 western hemlock seedlings at the Nursery. Benomyl, at 1 lb product/100 gal/acre applied at 10-to 14-day intervals throughout the growing season, is registered for control of Phomopsis-caused diseases, but the effectiveness of the fungicide against P. occulta has not been previously tested. Humboldt Nursery uses benomyl, but the efficacy of the treatment has been questionable. The increase in disease, along with the cost of benomyl and recommended frequency of application, caused the Nursery to request a Forest Pest Management evaluation of alternative fungicide treatments.

The objective of this study was to evaluate alternative fungicides for control of Phomopsis canker of Douglas-fir at Humboldt Nursery, through in vitro laboratory screening and field trials.

PROCEDURE

Laboratory Screening

An isolate of P. occulta obtained from 2-0 Douglas-fir seedlings at Humboldt Nursery was used. The isolate was morphologically identical to other Douglas-fir isolates and to isolates obtained from western hemlock.

A total of 16 fungicides were initially selected (Table 1) to determine their effect on spore germination. Fungicides for screening were recommended and supplied by Dr. A. H. McCain, Extension Plant Pathologist, Univeristy of California at Berkeley. Three concentrations (1, 10 and 100 ppm; active ingredient) of each fungicide were incorporated into 2% water agar (2 g agar per 100 ml water). Spores were obtained by adding ten ml of sterile water to each sporulating culture and brushing gently with a brush to dislodge spores. One ml of the spore suspension was placed onto the surface of each plate containing 2% water agar and incorporated fungicide. Four replications were made for each fungicide concentration and for a control (2% water agar without fungicide). After 24 hours incubation, percentage of germination was determined for each treatment. Development of germ tubes was followed over an 11-day period.

Table 1. Fungicides evaluated in vitro for control of Phomopsis canker of 2-0 Douglas-fir.

Common Name	Trade Name
benodamil	BAS 317F 50W
benomyl	Benlate
captan	Captan 50W
chlorothalonil	Bravo W-75
chlorothalonil	Bravo 500
diniconazole	Spotless (Chevron 779 12.5% W)
dithianon	Delan 75 WP
fenarimol	Rubigan 50W
iprodione	Chipco 26019
imazalil	Fungaflor 20% EC
mancozeb	Dithane M 45
penconazole	Topas (CGA 71818 10W)
prochloraz Mn complex	Sportak 50W
triadimefon	Bayleton 50WP
tri-basic copper sulfate	Tri-Basic
vinclozolin	Ornalin 50W

The seven fungicides that were most effective in inhibiting or reducing germination compared to the control were selected to determine their effect on mycelial growth. Three concentrations (1, 10 and 100 ppm; active ingredient)

were incorporated into potato dextrose agar (PDA). A 0.5 mm plug of mycelium of P. occulta was removed from the perimeter of an actively growing colony on PDA and placed on fungicide-amended PDA in a petri plate. Four replications were made for each fungicide concentration and for a control (PDA without fungicide). Radial growth of mycelium from each plug was measured after three days incubation.

Field Trials

Five fungicides were tested in field trials in 1985 and again in 1986. A randomized complete block design with six treatments (control; benomyl at 1 lb product/100 gal/acre; imazalil at 20 ounces product/100 gal/acre; diniconazole at 0.2 lbs product/100 gal/acre; chlorothalonil (Bravo 500) at 5 pints product/100 gal/acre; and mancozeb at 2 lbs product/100 gal/acre) was used. The six treatments were replicated 10 times down a bed of 2-0 Douglas-fir in 1985, and replicated 8 times in 1986. Each replicate covered 24 feet of bed (six, 4 X 4 ft plots), with each treatment assigned randomly in each replicate.

The 1985 trial was initiated in June. Fungicide treatments continued at monthly intervals through September, for a total of four applications. In 1986, monthly fungicide treatments began in May and continued through August (four treatments). Efficacy of treatments was determined by counting cankered seedlings monthly. After the final count of cankered seedlings, the total number of seedings in each plot was counted, and percentage of seedlings cankered in each treatment determined.

RESULTS AND DISCUSSION

Laboratory Screening

Germination percentages are presented in Table 2. Spores with emerging germ tubes after 24 hours incubation were considered germinated. With some treatments (imazalil, prochloraz, diniconazole, penconazole and benomyl in particular) however, germ tubes did not develop further or developed but became distorted. Therefore, the plates were observed for an additional 10 days, and germination rated on a 0 to 4 scale, with 0 = no germination, and 4 = germ tube length about equal to the control.

Germination ratings are presented in Table 3. Relative effectivess of the fungicides varied only slightly between the two methods of evaluation.

Table 2. Germination percentage of Phomopsis occulta spores after 24 hours on fungicide-amended media.

FUNGICIDE	CONCENTRATION		
	1 ppm	10 ppm	100 ppm
chlorothalonil (Bravo 500)	0	0	0
chlorothalonil (Bravo W-75)	71	0	0
imazalil	41*	43*	0
mancozeb	94	29	0
captan	92	89	0
dithianon	84	87	0
prochloraz	80	62*	31*
diniconazole	71	48*	28*
penconazole	87*	89*	67*
benomyl	95	88*	88*
benodamil	82	80	27*
tribasic copper sulfate	94	95	84*
fenarimol	90	88	86*
iprodione	91	90	89
vinclozolin	92	91	88
triadimefon	95	95	93
CONTROL	98		

* = germ tube inhibition or distortion.

Table 3. Average spore germination rating of Phomopsis occulta over an 11-day period on fungicide-amended media.

FUNGICIDE	CONCENTRATION		
	1 ppm	10 ppm	100 ppm
chlorothalonil (Bravo 500)	0[z]	0	0
chlorothalonil (Bravo W-75)	3	0	0
imazalil	1	1	0
mancozeb	2	1	0
prochloraz	2	1	1
diniconazole	3	1	1
penconazole	3	1	1
benomyl	4	1	1
dithianon	4	3	0
captan	4	4	1
tribasic copper sulfate	4	4	2
fenarimol	4	4	2
benodamil	4	4	3
vinclozolin	4	4	3
iprodione	4	4	4
triadimefon	4	4	4
CONTROL	4		

[z] 0 = no spore germination.
1 = germ tube development, but growth restricted or budding only.
2 = germ tube length less than 1/4 of control.
3 = germ tube length greater than 1/4, but not equal to control.
4 = germ tube length about equal to control.

Seven of the more effective fungicides in preventing or reducing spore germination were selected to determine their effect on mycelial growth. Results are presented in Table 4. After 11 days, no growth had occurred on benomyl-amended PDA at 1, 10 and 100 ppm, on imazalil-amended and procloraz-amended PDA at 10 and 100 ppm, and on diniconazole and Bravo 500-amended media at 100 ppm.

To determine if the fungicides were fungitoxic or fungistatic, plugs where no mycelial growth had occurred into the fungicide-amended PDA were removed, placed on PDA, and examined 7 days later. Results are presented in Table 5.

Table 4. Average radial growth of Phomopsis occulta after three days on fungicide-amended potato dextrose agar.

FUNGICIDE	RADIAL GROWTH (mm) AT THREE CONCENTRATIONS		
	1 ppm	10 ppm	100 ppm
benomyl	0	0	0
imazalil	0	0	0
prochloraz	0	0	0
diniconazole	1.1[z]	0	0
chlorothalonil (Bravo 500)	2.2	0	0
chlorothalonil (Bravo W-75)	15.5	1.7	0
mancozeb	24.0	2.7	0
CONTROL 27.5			

[z]Average of four replications.

Table 5. Mycelial growth of Phomopsis occulta on potato dextrose agar after 11 days on fungicide-amended potato dextrose agar.

FUNGICIDE	CONCENTRATION		
	1 ppm	10 ppm	100 ppm
benomyl	-[z]	-	-
imazalil	-	-	-
prochloraz	+	-	-
diniconazole	+	-	-
chlorothalonil (Bravo 500)	+	+	-

[z]+ = growth
- = no growth

Benomyl and imazalil were fungitoxic at all concentrations. Prochloraz and diniconazole were not fungitoxic at 1ppm, but were at 10 and 100 ppm, with mycelial growth occurring after the plug was removed from the fungicide-amended media. Chlorothalonil was fungitoxic only at 100 ppm.

Field Trials

Based on the laboratory assays, five fungicides (Bravo 500 formulation of chlorothalonil, imazalil, diniconazole, benomyl, and mancozeb) were selected for field trials. Although prochloraz was effective in laboratory assays, the fungicide was not available from the manufacturer at the time of the 1985 field trial.

Levels of disease incidence during the 1985 trial were not sufficient to compare treatments. Cankered seedlings in plots ranged from less than 1% to about 3%.

Results of the 1986 trial are presented in Table 6. All five fungicides tested gave significantly better control of Phomopsis canker than no treatment. Benomyl, the fungicide registered for Phomopsis control, was more efficacious than mancozeb or imazalil, but was not as effective as diniconazole or chlorothalonil.

Table 6. Percentage of 2-0 Douglas-fir seedlings cankered as a result of Phomopsis occulta infection at the Humboldt Nursery.

TREATMENT	PERCENT CANKERED
diniconazole	1.9 a[1]
chlorothalonil	2.1 a
benomyl	4.0 b
mancozeb	5.1 c
imazalil	5.3 c
control	9.1 d

[1]Means followed by the same letter are not significantly different at the 1% level, according to Duncan's Multiple Range Test. Means are the average number of seedlings cankered in eight replicate plots.

The label for benomyl suggests treatment at 10-to 14-day intervals. Because we used 28-to 30-day intervals, the fungicide may be more effective if used according to label recommendations. Chlorothalonil is registered for use on conifer seedlings in nurseries and, based on results of these studies, could be used in place of benomyl for control of Phomopsis canker. Diniconazole has some systemic properties and therefore may be more effective than chlorothalonil or benomyl in preventing secondary infections during rainy periods. Diniconazole is not currently registered for use in nurseries in California.

CONCLUSIONS

Benomyl, the fungicide registered for Phomopsis control, was efficacious under the conditions of this study. Chlorothalonil and diniconazole were more effective than benomyl. Chlorothalonil is registered for use on conifers in nurseries, and could be used (5 pints product/100 gal/acre, applied at monthly intervals throughout the growing season) as a replacement for benomyl.

Although fungicide treatments for control of Phomopsis canker were effective under the conditions of this trial, results should be interpreted with caution because of the low incidence of disease during the two seasons of field testing. Incidence of Phomopsis canker in non-treated portions of the test bed in the 1986 trial was low (9.1%), with little variation among the eight replications. During years more favorable for disease development, the relative efficacy of the fungicides evaluated in relation to each other may vary.

Most of the cankers recorded over two years had caused dieback of lateral tips, with only about 10% of these infections resulting in death of the seedlings or cull due to killing of terminals. This, along with the low levels of overall infection in test beds and production beds, suggests that no fungicide treatment the past two growing seasons may have been more economical than treatment with preventive fungicides. Observations in 1986 suggested that disease levels may have been lower than in previous years because seedlings were not top-pruned, a practice that creates wounds for infection.